1.

(a) (i) higher temperature increases kinetic energy of enzyme;
more (successful) collisions;
more enzyme – substrate complexes formed;
faster production of maltose / the product / faster rate of reaction;
reaches end point quicker;
enzyme at 25°C converts all of substrate eventually;

[4 max]

(ii) graph showing reverse shape; [1]
reaches zero at between 6-8 minutes; [1]

(iii) pH / enzyme / starch concentration; [1]

(b) (i) 1 [1]

(ii) at high temperatures enzyme molecules vibrate vigorously;
hydrogen bonds broken;
shape of active site changed;
enzyme substrate complexes not formed / substrate cannot fit into active site;
denatures;

[3 max]

(iii) only four tested;
only shown to be effective against milk protein (not other proteins);
no results for starch / carbohydrate / fat;
powder may not digest these;

[2 max]

[Total 13 marks]

2.

(a) (i) A competitive [1]

B non competitive [1]

(ii) A [1]

(b) (i) 5°C kinetic energy is low / few collisions between the (active site) of the enzyme and the substrate; [1]
Allow: ref. to increasing temp and kinetic energy i.e. assume 0 to 5°
70°C the hydrogen bonds are broken (as vibrations are strong) / active site of the enzyme is denatured/ fewer ES complexes formed due to denaturation. [1]

(ii) A. activity of immobilised enzyme is greater between 0°C and 40°C or at lower temperatures/ rate of reaction greater

B. optimum temperature of IE covers a wider range / 40°C - 50°C

C. above 40°C the free enzyme begins to denature whereas the IE starts to denature at 50°C

D. IE is more active at all temperatures except 40°C

E. free enzyme is (completely) denatured at 70°C IE is completely denatured at 80°C

(any three) [3]

(iii) The shape of the enzyme / 3-D structure is maintained or it is stabilised – molecular movement is 'reduced' [1]
(not: shielded/protected enzyme)

(iv) Detection of blood sugar / testing blood sugar (in diabetics)
Allow : lactose free milk/Clinistix qual
(not: diabetics/ biosensor) [1]

[Total 10 marks]

3.

(a) (i) Showing, 1 O and 2 H s removed.
Elimination of water, stated.
Molecules joined by oxygen bridge. [3]

(ii) Maltose (not: disaccharide). [1]

(iii) Water. [1]

(iv) Condensation. [1]

(b) (i) Joining together sub units / monomers /repeating units/ residues
(to make a larger molecule) [1]
(not: joining molecules into a chain/ specific example)

(ii) Correct axes – iron sulphate concentrate on horizontal, both labelled and units given.

Suitable scale using at least half available space;

plots visible and clear line correct shape. [3]
(not: extrapolation/line of best fit)

(iii) 0.9mM (allow: between 0.7 and 0.9mM). [1]

(iv) 60 – 5.2 = 54.8 / 60 x 100 = 91.3(%) (allow: 91)

(2 for correct answer 1 for correct working but wrong answer.) [2]

(v) Inhibitor competes with substrate (to bind with active site);

inhibitor binds to/fits into active site;

with inhibitor bound substrate is unable to bind/less E-S complexes;

inhibitor same/complementary shape as substrate;

the greater the concentration of substrate the less inhibition / ra / owtte

(Any 3) [3]

(vi) (Add iron sulphate to toothpaste / mouthwash / sugary drinks.)
to prevent formation of plaque / tooth decay. [1]

[Total 17 marks]

4.

(a) (i) alginate beads / gel membrane / meshwork of inert material / cellulose (not: entrapment unqualified) [1]

(ii) product easily recovered/not contaminated by enzyme;
so cheaper to use;
greater stability;
despite variations in/higher temperature / pH;
enzyme easily removed / added;
can control rate.
more than one enzyme can be used [3]

(b) (i) allows urea to pass through;
prevents passage of blood cells / other molecules/solutes;
so they can't affect results / enzyme / reduce enzyme activity. [2 max]

(ii) absorb/ref. ammonium ions;
converts into an electrical signal / changes chemical to electrical signal;
to record levels of urea. [2]

(c) increased temperature increases enzyme activity/rate of reaction;
more ammonium ions formed;
greater electrical current generated;
reference fair testing. [2 max]

(d) diabetes. [1]

[Total 11 marks]

5.

(a) (i) nitrogen containing part; [1]

(ii) arrow pointing to glycosidic bond; [1]

(iii) hydrolysis; [1]

(iv) hydroxyl groups point outwards;

link with neighbouring chains;

via hydrogen bonding;

to form microfibrils;

strong structure because of large number of hydrogen bonds;

chains associate in groups / fibres formed;

beta glucose units.
ref. alternating rotation [3 max]

(b) (i) tertiary; [1]

(ii) links between different parts of polypeptide chains;

produces a specific shape for the molecule / lysozyme;

reference to active site;

complementary to substrate;

allows enzyme – substrate complexes to form; [3 max]

(c) (i) mass/volume of tissue/sample; (not: amount/size)

concentration of hydrogen peroxide;

same time intervals between measurements;

equal volumes of hydrogen peroxide used;

pH;

temperature. [2 max]

(ii) most metabolically active;

produces most hydrogen peroxide;

needs to be broken down because of toxicity; [2 max]

[Total 14 marks]

6.

(a) (i) At higher temperature/60° enzyme/substrate has more kinetic energy/vibrates more; (not: ref. movement) 3

More Enzyme substrate complexes formed/ more successful collisions;

More product formed/greater rate of reaction.

(ii) At 60ºC enzyme reacts rapidly; 3

(Gradual) denaturation of enzyme occurs or description;

All substrate not reacted;

(b) All substrate converted to product. 1

(not: active sites full)

(c) Lower temperature, less kinetic energy/fewer vibrations; (3 max)

Fewer enzyme substrate complexes formed/fewer successful collisions;

Some substrate remains after 60 minutes; (not: reaction has not ended)

Maximum product formation not yet achieved.

10 MARKS

7.

(a) (i) Can be re-used; (2 max)

Greater stability;

Despite variations in temperature/pH;

Easy to remove product/product not contaminated with enzyme;

More than 1 enzyme can be used/enzymes added or removed easily.

Can be used in a continuous production system

(ii) Colour change only/can only indicate if its present or absent; 2

Subjective nature of judgement of colour/qualitative rather than quantitative.

(b) (i) Measures metabolite/named substance; 2

By converting chemical signal/energy into an electrical signal/energy.

(ii) Combines with substrate/glucose; (2 max)

At active site;

To produce product.

(iii) Glucose from blood diffuses into gel; (4 max)

Acted on by glucose oxidase;

Amount of product released proportional to glucose concentration;

Electrode activated by product;

Generates electrical potential/signal;

Size of potential directly proportional to mass of product.

12 MARKS

8.

(a) capable of immobilisation/fixed to inert matrix or named;
Stable/able to withstand changes in temperature or pH;
specific to test or substrate;
(not: ref. turn over number)
2 max

(b) allows glucose through; (not: ref. small molecules)
prevents passage of other molecules/solutes
(not: ref. substances)
2

(c) glucose broken down by enzyme;
Products/oxygen affect/detected by electrode;
(not: measured by)
electric signal generated/chemical to electrical;
greater conc. glucose the greater the signal;
2 max

(d) enzyme activity/ rate of diffusion of glucose affected;
change rate of reaction;
unreliable result;
(not: ref. enzyme denaturation/fair experiment/control/false reading/confidence)
2 max

(Total 8 marks)

9.

(a) $\frac{5.8}{0.5}$
11.6 cm^3 min^{-1};
(allow: 5.8/30 x 60) correct answer + units =2;
correct answer - units =1; incorrect answer, correct working = 1
2

(b) Maximum/higher concentration of substrate;
all active sites occupied;
(not: ref. unoccupied at start)
2

(c) (i) increase in rate from 20 - 100°C/up to 100°C;
fall from 100 - 130°C;
increase in kinetic energy;
molecules move faster; (not: more)
More successful collisions/more enzyme-substrate complexes formed;
up to optimum; (not: 100°C unqualified)
above optimum increased vibrations;
hydrogen bonds break;
Loss/change of shape of active site; (not: ref. enzyme)
denature;
6 max

(ii) enzymes have different optimum temperatures/
human amylase has optimum of 37°C, bacterial 100°C;
1
human amylase denatures at a lower temperature;
1

(Total 12 marks)

10.

(a)	(i)	0.26	1
	(ii)	concentration of substrate NOT amount/ availability of Active sites	1
(b)	(i)	Less/low kinetic energy ;	1
		fewer successful collisions/ {enzyme substrate/ES} complexes formed/ ORA	1
	(ii)	enzymes denatured/ alteration in tertiary structure/ 3D structure; breaking of H/ hydrogen bonds; NOT disulphide active site altered/ active site denatured; substrate cannot bind/ less enzyme substrate complexes formed (any three)	3
(c)	(i)	Must be a curve starting at origin and may meet 30°C line but not levelling off	1
	(ii)	{shape/structure} of inhibitor similar to substrate/complementary to active site; {Fits/ fills/ bonds/ attaches} to active site/ competes for active site; (As it has a similar shape to the substrate it competes for the active site = 2 marks.) At higher substrate concentration there is a greater chance of Enzyme substrate complexes forming / effect of inhibitor is diminished/ the substrate outcompetes the inhibitor/ ORA	3

(Total 11 marks)

11.

(a)	(i)	Molecule of water (drawn with arrow towards the O atom of the glycosidic bond); NOT water going out Monosaccharides drawn with –OH groups in correct position on C1 and C4 (involved in bond);	2
	(ii)	Hydrolysis; NOT hydrolysation (ignore reference to acid)	1
	(iii)	Glycosidic;	1
	(iv)	Glucose and galactose; ignore alpha/ beta	1
(b)	(i)	An enzyme that has been fixed to an inert {matrix/support/ substance};	1
	(ii)	The enzyme can easily be recovered/ reused; The product is free from contamination; Enzyme is {stable at / tolerates/ withstand} higher temperatures/denatures at a higher temperature/ functions over a wide range of pH; NOT wider range of temperature alone Several enzymes with differing optima can be used at the same time; More control over the reaction/enzymes easily added or removed/ can be used in a continuous process;	Max 2

(c) (i) Heat with Benedict's solution/reagent; 2
NOT warm/ water bath/ ref to acid
Blue to{red/ orange/ green/ yellow/ brown};

(ii) Instrument/equipment that can detect a specific 1
molecule/metabolite (in a mixture of molecules/bodily fluid).

(iii) Any one from: 1
The biosensor would give quantitative data/
it would detect {a particular product/glucose/galactose}/
Can detect even at {very low concentrations/ small volumes};

(d)

1. (The concentration of reducing sugars) would decrease; Max 4
2. {Lactose/ substrate} concentration is lower (in the sour milk);
3. Lactic acid lowers the pH;
4. Enzyme would be inactivated/denatured;
5. Hydrogen/ ionic bonds (maintaining the 3D shape) would break;
6. This will change the shape/charge of the active site (of lactase);
7. Fewer enzyme-substrate complexes would be formed/fewer successful collisions;
8. Benedicts would remain {blue/ change to {orange/ yellow/ green/ brown}/ negative}

Question 6 Total **[16]**

12.

(a)	(i)	Lock and key;	1
	(ii)	Theory 1/ induced fit;	1
(b)		Enzyme substrate complex; NOT ESC/ ES complex	1
(c)		Lower the activation energy/eq;	1
(d)		Enzyme/ active site is unchanged/can be re-used; NOT active sites are a specific shape unqualified	1
(e)		Temperature (not heat); pH; NOT acidity Enzyme concentration; Substrate concentration; NOT amount	3
(f)		Intracellular: inside the cell + Extracellular:outside the cell; NOT inside body	1
		Question 2 total	**[9]**

13.

(a)		Causes change in shape of enzyme/active site; So substrate no longer fits into active site; {No/ fewer} enzyme substrate complexes;	2 max
(b)		{(Insoluble) enzymes/ (enzyme) aggregates} cannot pass through the filter/ ORA; So the product is uncontaminated with enzymes/ ORA;	2
(c)		Can tolerate {higher temperatures/greater range of pHs}; NOT range of temperatures Easily recovered for reuse/ enzymes stay in aggregates/ reused qualified/ uncontaminated product/ separated from product; NOT reused unqualified/ enzymes reused Several enzymes can be used together; Easy addition/removal of enzymes;	3 max
(d)		Any one from : Gel capsule/alginate beads/ gel beads; cellulose fibres; gel membrane; porous glass beads; NOT inert matrix unqualified/ encapsulation unqualified	1 max
		Question 6 Total	**[8]**

14.

(a)	(i)	Activation energy;	1
	(ii)	Line starting and finishing at the same point but with a lower activation energy;	1
(b)		The <u>active site</u> (of succinate dehydrogenase) has a <u>specific shape</u>; Succinate has a <u>complementary</u> shape; (and therefore) {fits/ binds/ bonds to} into the active site; NOT attaches	Max 2
(c)	(i)	I The concentration of succinate/ substrate; II As the concentration of the {succinate/substrate} increases {the rate of reaction/production of fumarate increases};	1 1
	(ii)	The concentration of succinate dehydrogenase/ enzyme; all of its active sites are occupied (at any given moment);	2
(d)	(i)	Malonate has a similar {shape/structure} to {succinate/ substrate} / malonate has a complementary {shape/structure} the active site; NOT same shape Malonate {binds/ competes} to the active site; Prevents succinate binding / fewer enzyme-substrate complexes are formed; (MP3 must be in context of competitive inhibition)	3
	(ii)	Curve rising at a lower rate and plateaus at the max rate at a higher concentration; Accept max rate may not be reached	1
		Question 4 Total	**[12]**

15.

(a) (i) Allows the glucose molecules to pass through (to the enzyme layer); — 2 max
Prevents the passage of other solutes ;
so they can't {affect results / affect enzyme / reduce enzyme activity};

(ii) glucose broken down by enzyme; — 3 max
the {hydrogen peroxide/oxygen} is {detected/absorbed} by electrode;
an electric signal is generated/ changes chemical to electrical signal;
the greater the concentration of {glucose/hydrogen peroxide/oxygen}
the greater the signal;

(b) (i) The enzyme converts glucose into it's isomer fructose / glucose and fructose are isomers; — 1

(ii) Add Biuret solution / sodium hydroxide solution & copper sulphate; — 2
(reject if reference to heat)
The solution would remain blue / no colour change would occur;

(iii) can be re-used; — 2 max
has greater stability/denature at higher temperatures;
can catalyse reactions/greater stability over a wider range of pH;
More than one enzyme can be used/enzymes added or removed easily/ greater control over process/ can be used in a continuous process;
(Reference to cost is neutral)

Question 6 Total — **[10]**

16.

(a)	Temperature from 0-30. Peak between 35 and 40. graph finishing at 55-65.	1 1 1
(b) (i)	As temperature rises more collisions occur / molecules have greater (kinetic) energy / move faster; (1) between active site and substrate; (not: enzyme) (1) so more enzyme-substrate complexes are formed / rate of reaction increases. (1) Any 2.	2

(ii)	As temperature rises (above optimum), bonds in the protein / enzyme are broken; (1) This alters the shape of the active site / denatures the enzyme / protein so the substrate no longer fits; (1) so rate of reaction slows. (1) reaction stops at 55-65°C as all enzyme active sites / molecules are destroyed / denatured. (1) Any 3.	3
(c)	pH; (1) enzyme concentration; (not: amount) (1) substrate concentration. (no double penalty) (1) Any 2.	2
		[10]

17.

(a)	Immobilised enzymes are enzymes that are enmeshed in / attached to an (inert) solid support / Enzymes stabilised on a (gel) membrane / (Accept enzymes in alginate beads.)	1
(b)	The substrate / glucose is turned into a product. The glucose attaches to the enzyme (and product is formed.) / reference to an active site	2
(c)	Glucose oxidase electrode detects the glucose / Transduction into electrical impulses / The current produced can be read on the scale / The product causes a change in the potential difference which is measured by an electrode.	1
(d) (i)	To regulate / maintain pH / enzymes can only work at a specific pH.	1
(ii)	Temperatiure	1
(e)	 Y anywhere except inside	2
(f)	X – Similar shape to that of the normal substrate and so it fits into and blocks the active site. (not: competes unqualified.) Y – Binds to the enzyme outside of the active site and changes the shape of the enzyme / active site / precipitate the protein.	2
(g)	Malonate / malonic acid / antibiotics / sulphonamide drugs.	1 [11]

18.

a	i	competitive;	1
	ii	fits into (part of) active site;	
		prevents substrate from entering/blocks site;	2
b	i	curve starts to rise above 5°C;	
		optimum at 40°C;	
		steep decline to zero rate at 50°C;	
		rise is concave;	
		Axes correct	MAX 4
	ii	kinetic energy low/molecules move slowly/	
		has low/less chance of collision with enzyme;	1
	iii	bonds break / shape of active site distorted/changed/loss of tertiary structure	1

19.

a	i	Enzyme held/stabilised in inert support/matrix or example;	1
	ii	can be reused;	
		does not contaminate product;	
		faster/more controllable/efficient process;	
		more resistent to changes in temperature/pH	MAX 2
b	i	300(mg/min at 4.5)	
		200(mg/min at 2.5);	1
		(300-200=100mg x 60) =6000 OR 6g;(per hour)	1
		units g or mg (answer per min = 2 max)	1
	ii	All active sites in use/all enzyme molecules saturated;	
		Maximum/optimum rate of conversion/ fructose production reached/max turnover number (amount of enzyme becomes a limiting factor, 1max)	2

20.

(a) (A substance) which speeds up the rate of a chemical reaction (1)

(b) Activation energy/energy required (1)

(c) (Increasing conc. of malonic acid) reduces the rate of reaction/increases inhibition (1)

(d) Competitive (1)

(e) The malonic acid/inhibitor has similar structure to succinic/substrate (could be show in diagram) (1)
Competes with substrate for active sites/binds with active site (1)
The more sites occupied by inhibitor the fewer there are for substrate/ decrease in enzyme - substrate complexes. (1)
(allow consequential error if non-competitive, inhibitor binds to site other than active site(1), changes shape of active site (1) substrate no longer fits (1)

Page Break

(f) Use a buffer/add acid and alkali with monitoring. (1)

(g) Replicate conditions/repeat experiment (1)
Using boiled (and cooled)/denatured extract (1)
(not: ref. to water/leave enzyme out)

Total = 10 marks

21.

(a) Linear scales (both) + good use of paper. (over 50% both directions) (1)

pH along X axis % up Y axis. Correct positions + axes labels on graph including % or proportion. (1)

Correct plots (-1 for each incorrect plot). (2)

Smooth lines drawn for curves. (1)

(b) (i) O / no activity (1)

(ii) Peak at 2 (± ½ pH) (1)

Narrow range (0 → 5) (1)

(c) Alter (the shape) of the active site / destroyed / damaged;

(Extreme) changes will denature enzyme (allow: changes 3D shape if very clear);

Changes in pH alter the ionic / electrostatic charges (of the acid and base groups);

Inactive / lower activity with a small change.

Any three (3)

(d) Temperature (not: heat / room temperature);

Enzyme concentration (not: amount / volume);

Substrate concentrations (not: amount / volume / concentration solute);

Time.

Any two (2)

(e) Buffers
(1)

Total 14 marks

22.

(a)	(i)	Lipase	1
	(ii)	Breaking of a bond (allow: large to small molecules) Chemical insertion (not: addition) Of water	2
	(iii)	pH reduced/more acidic	1
		Lipid hydrolysed to fatty acids/ (Fatty acids) cause low/acidic pH	1
(b)	(i)	Start with maximum marks and deduct one at a time for the following errors: Incorrect plot Poorly drawn curve (allow: ruled lines) Inappropriate scales (insufficient use of graph paper) Axes not labelled/no units	3
	(ii)	Rate remains constant/can't increase	1
		Substrate/lipid occupies active site/at lower concentrations of lipid not all active sites occupied/ at 0.7% lipid concentration all sites occupied	
		Extra lipid can't access sites (not: limiting factor unqualified)	1
	(iii)	Steeper curve (up to 0.7%)	1
			[11]

23.

(a)	active site		1
(b)	lock and key/induced fit		1
(c)	hydrogen/sulphur/peptide/ionic bond/van der Waals/disulphide bridges (not: covalent)		2
(d)	A		1
(e)	ATP		1
(f)	(i)	anticodon	1
	(ii)	determines specificity/order of amino acid	1
			[8]

24.

(a)	Temperature and pH		2
(b)	(i)	The concentration of substrate is in excess/active sites occupied/enzyme limiting factor	1
	(ii)	substrate has been used; its concentration falls	1
(c)	starting point of line at 50% or 100% mark		1
	duration of horizontal line 12 minutes		1
	zero value at 20 minutes		1
(d)	(i)	an enzyme absorbed on an (inert) support/example	1
	(ii)	easier to control reaction/reusable/no need to separate from product/resistant to change (e.g. pH) rate of reaction greater/stabilised qualified. (Any 2) (not: cheaper/quicker)	2
	(iii)	fermentation/fruit juice manufacture/pregnancy kits/urea or sugar in blood/reduction of lactose in milk/biosensors qualified. (not: ref. to baby foods unqualified/cheap/pollution)	1
			[11]

25.

(a)	Temperature and pH		2
(b)	(i)	The concentration of substrate is in excess/active sites occupied/enzyme limiting factor	1
	(ii)	substrate has been used; its concentration falls	1
(c)	starting point of line at 50% or 100% mark		1
	duration of horizontal line 12 minutes		1
	zero value at 20 minutes		1
(d)	(i)	an enzyme absorbed on an (inert) support/example	1
	(ii)	easier to control reaction/reusable/no need to separate from product/resistant to change (e.g. pH) rate of reaction greater/stabilised qualified. (Any 2) (not: cheaper/quicker)	2
	(iii)	fermentation/fruit juice manufacture/pregnancy kits/urea or sugar in blood/reduction of lactose in milk/biosensors qualified. (not: ref. to baby foods unqualified/cheap/pollution)	1

[11]

26.

Immobilised enzyme mark scheme

(a) (i) 2(mm)
(because) it gives the highest percentage of product 1
(not: converse)

(ii) smaller beads give a larger total area (exposed to the milk/substrate);
(not: smaller beads have large surface area)

or

smaller beads will pack more closely (so there is more enzyme:subtrate);

the milk will have longer contact/flow more slowly around small, close packed beads;

or

gives more time for the enzyme and subtrate to come into contact. 2

(iii) Percentage product would increase/get more product;

More time for enzyme-substrate complexes to occur/ for reaction. 1

(iv) temperature/milk type/source (not: pH) 1

(b) glucose and galactose 1

(c) Can tolerate/more stable in a wider range of condition/temps/pH qualified;

Product not contaminated with enzyme/pure product/easily reused qualified;

Several enzymes (requiring different conditions) may be used together;

Enzymes easily added/removed = more control. 2
(allow: ref. to continuous production qualified,
not: ref. to cost)

Total 8 marks

27.

(a)	Increased conc increased rate of reaction.	1
(b)	5.5; (allow : 5.6)	1
	Fig. from graph (1.1 to 1.2)	1
(c)	Denature/boil enzyme (not: no enzyme)	1
(d)	Enzyme has a pH optimum/does not work in acid pH (allow: denatured not: destroyed/inhibited)	1
(e)	Faster (colour change) at start as temperature increases;	1
	No further colour change after enzyme denatured.	1
(f)	rate drops/stops if all active sites blocked;	1
	competes with normal substrate for active sites.	1
(g)	Biuret test/NaOH or any alkali plus $CuSO_4$;	1
	Purple/violet ring/colour develops if protein. (linked marks)	1 [11]

28.

(a)	pH; temperature; enzyme concentration;	3
(b)	(i) A;	1
	(ii) increased substrate concentration reduces effect/allows rate to reach maximum eventually/AW;	1
	(iii) similar shape/conformation to active site/substrate; attaches to/occupies active site; (not: competes with active site) blocks site/prevents substrate from binding; no enzyme-substrate complexes formed/substrate not broken down; no products produced/released; reversible; (not: stops or slows reaction)	3 Max
		[8]

29.

(a)	(i)	$12cm^3\ O_2\ min^{-1}$	1 mark
	(ii)	Increase of 11.75 or 48 times	1 mark
(b)	(i)	Concentration of enzyme (Not amount)	1 mark
	(ii)	Concentration of substrate (Not amount)	1 mark
(c)	(i)	'Small quantity', so line more than half way up to original line and same shape. Line above 5 on y axis.	1 mark
	(ii)	Lowered position because low concentration alters the structure of some of the enzyme molecules rendering them non-functional (lowers number of active sites ok, but lowers the amount of enzyme is not acceptable.)	1 mark
	(iii)	Non-competitive because inhibitor molecule does not compete with substrate for active site.	1 mark
		Attaches elsewhere on the enzyme, distorting the active site.	1 mark
(d)		Breaks tissue/cell walls to release enzyme/increases surface area for substrate contact.	1 mark
(e)		pH changes may otherwise occur during the experiment/	1 mark
		which alter the shape of the protein and affects the efficiency of the active sites.	1 mark
(f)		Reaction may take up or produce heat, causing changes in rate./Fluctuations in external temperature have same effect.	1 mark
(g)		Denaturation/breakdown of active site.	1 mark

Total 13 marks

30.

(a) (i) protein; [1]

(ii) correct part labelled/circled; [1]

(iii) complementary shape to glucose/ similar shape to substrate and active site;
substrate/glucose will fit into/binds with (active site)/ enzyme;
(not: competes)
can form enzyme substrate complex;
glucose can't fit anywhere else; Max [2]

(b) complementary/similar shape to active site;
(not: ref. to substrate)
enters active site/ occupies active site;
blocks active site;
(not: competes/ joins active site)
no glucose can enter;
less/ no enzyme substrate complex formed/ fewer products formed;
glucose not metabolised/no products formed/ metabolised more slowly;
competitive inhibitor; Max [4]

(c) ethanol acts as inhibitor;
competitive;
meths can't be converted to formaldehyde/converted more slowly;
no/reduced toxic effects Max [3]

Total [11]

31.

(a) (i) Transport against a concentration gradient and requiring energy input. (1)

(ii) The order/sequence in which amino acids are linked in the protein. (1)
(not : chain of amino acids/ref to bends)

(iii) The compound formed by substrate joining the active site. (1)

(iv) Enzyme trapped in/adsorbed on an inert carrier/gel matrix. (1)

Total 4

32.

(a) (i) pH (1)

(ii) (Add the same volume of) buffer solution to each tube. (1)

(iii) Changing pH might change the active site / distort shape. (not : denature) (1)

(b) 360-300 = 60 60/5 = 12 mg min^{-1} (must give units) (1)

(c) (i) Less (1)

(ii) Enzyme working at full capacity/all active sites in use/

spare/excess substrate molecules/enzyme is limiting factor /

rate of reaction is constant for 15 mins (any two) (2)

(d) The starting value will still be 24 (no increase in active sites) (1)

The first six boxes should contain 24 (double the concentration of substrate means it will take double the time to come down to enzyme concentration) (1)

The seventh box should be 12 and the last one zero (The fall will be just as rapid as in the first experiment once the concentrations have reached equality) (1)

[If the calculation in (b) is incorrect, their incorrect value can be accepted in (d) provided it is less than 24.]

Total 10

33.

(a) (i) glycerol;

3 fatty acids
(labels required) [max 2]

(ii) ester [1]

(iii) phospholipid has phosphoric acid;

phospholipid 2 fatty acids;

phospholipid hydrophobic tails and hydrophilic head/polar molecule;
[max 2]

(b) fats have higher energy value (animal will not be as heavy if

storing same energy values as carbohydrate.) [1]
(not: ref. other functions/more efficient)

(c) saturated more hydrogen;
Saturated solid at room temperature and unsaturated are liquid;
unsaturated double bonds and saturated single/no double bonds;
unsaturated kinky/bent tails and saturated straight.
[max 2]

(d) (i) make contents of tubes alkaline/raise pH [1]
(not: ref. optimum pH)

(ii) red (allow: green)

(iii) lipase/enzyme hydrolyses/splits fats;

fatty acids make contents acidic/lowering pH [2]

(iv) (heating has) changed shape of active sites;
(not: destroy)
denatured enzyme/ bonds broken;
substrate no longer fits/complementary/ no enzyme-substrate complex forms.
no fatty acids produced [2]
(not: ref. product/no digestion)

Essays
1.

(b)	A.	Enzyme molecules that are fixed / bound / Trapped (not: immobilised/do not move)	[1]
	B.	to an inert Matrix/alginate bead.	[1]
	C.	They are more stable at higher temperatures (therefore reaction rates may be faster by using higher temps.)	[1]
	D.	They can tolerate wider range of pH.	[1]
	E.	They are more easily recovered for re-use/separated from product.	[1]
	F.	Several enzymes with different pH or temp. optima may be used at one time.	[1]
	G.	Reaction can be more easily controlled by adding or removing enzymes.	[1]
	H.	They are specific so can select one type of molecule in a mixture.	[1]
	I.	So can be used for rapid detection of biologically important molecules.	[1]
	J .	They can also accurately measure the quantities present / are sensitive.	[1]
	K.	Used in medical diagnosis / named condition eg diabetes.	[1]
	L.	And environmental monitoring,	[1]
	M.	Description of mechanism, some use a transducer to generate an electrical impulse that can be measured with a meter.	[1]
	N.	eg. Blood sugar meter as used by diabetics / AVP.	[1]
	O.	AVP/ easier to make pure product (not contaminated by enzyme.)	

[Total 10 marks]

2.

(b) (i)

A. two types, competitive and non-competitive;

B. both types of inhibitors reduce rate of reaction;

C. competitive inhibitor complementary to active site / structurally similar to substrate;

D. competes with substrate for active site of enzyme;

E. blocks active site/prevents substrate from binding to active site

F. fewer/ no enzyme substrate complexes formed;

G. increase substrate concentration reduces effect of inhibitor;

H. non-competitive binds away from active site/ binds at allosteric site

I. changes shape/conformation of enzyme molecule;

J. shape/conformation of active site changed;

K. increasing substrate concentration has no effect on rate of reaction **[7]**

(ii)

L. enzymes tolerate wider range of conditions/temp/pH/thermostable/ Owtte (not: stable unqual)

M. enzyme easily reused;

N. several enzymes can be used together;

O. product not contaminated / easier purification of product

P. greater central of reaction achieved/ enzymes easily added or Removed qual. **[3]**

(Total 10 Marks)

3.

(a) A. enzyme (molecules) {fixed/ bound/ trapped} in an {inert support/ matrix}

B alginate beads/ gel membrane, /adsorbed (NOT absorbed)onto nylon/ gel capsule/ cellulose

C Product not contaminated

D reuse of enzymes/recovery/ easily separated.

E stable/ tolerate wider range of conditions

F for example pH, temperature/ higher temperatures than normal/ denatured at higher temperatures

G several enzymes can be used together/ with differing pH or temperature optima.

H rapid/ greater productivity

Biosensors

I accurate/ specific

J detect/sensitive to low concentrations/ clinistix

K used in diagnosis of diabetics/ diabetes

L {Biosensor/electrode probe} has a specific enzyme immobilised in a membrane/ glucose oxidase in context

M glucose diffuses into the immobilised enzyme layer/ through selectively permeable membrane

N (enzyme together with transducer) produces an electrical signal in response to substrate transformation/ chemical to electrical signal

O size of signal proportional to concentration of product/ substrate

(Any 10 out of 15 points)

4.

(b)	A	Temperature;	1
	B	description of (exponential) increase to optimum / maximum / certain temperature then (sudden) decline / sketch graph showing;	1
	C	Increasing temperature increases rate because of increased energy / moving molecules faster / kinetic energy / ORA;	1
	D	{Increasing frequency of / more / more likely} successful collisions / Enzyme Substrate Complexes forming / ORA;	1
	E	pH;	1
	F	description of optimum pH and declining activity further from optimum in both directions / sketch graph / optimum pH and narrow range;	1
		(Award G, H, I, J in context for Temp and/or pH)	
	G	(3D) shape of active site changes;	1
	H	Changing away from optimum affects bonds holding tertiary structure / structure of enzyme molecules;	1
	I	Correct reference to hydrogen / covalent / ionic bonds; NOT disulphide / peptide	1
	J	Substrates do not fit into active site / is not complementary (so rate reduced);	1
	K	Substrate concentration; NOT amount;	1
	L	Enzyme concentration; NOT amount;	1
		(Award M,N, O in context for Enzyme conc and/or Substrate conc)	
	M	Activity increases up to maximum when it levels off / sketch graph showing / ORA;	1
	N	Increasing substrate / enzyme conc. increases number of active sites occupied / Enzyme Substrate complexes / successful collisions / ORA;	1
	O	Maximum rate when all active sites occupied / saturated correct reference to limiting factors;	1

5.

(a) **Describe and explain the effect of inhibitors on enzyme action.** **[10]**

enzymes

A Enzymes are globular proteins/<u>biological</u> catalysts;

B Active site (of the enzyme) has a specific 3D/ tertiary shape;

C lower activation energy of a reaction;

D Inhibitors reduce the rate of (an enzyme catalysed) reaction;

competitive

E Competitive inhibitors;

F Have a shape similar to the substrate/complementary to the active site; NOT same shape

G Fit/ bind into the active site;

H Prevent the substrate molecule entering the active site/block the active site;

I Max. rate of reaction can be achieved at higher substrate concentrations/ Increasing the concentration of the substrate reduces the effect of the inhibitor; allow correctly labelled graph

non-competitive

J Non-competitive inhibitors;

K Bind to the allosteric site/site other than the active site;

L Causes a change in the shape of the <u>active site</u>;

M Substrate can no longer fit into the <u>active site/ active site is no longer complementary</u>;

N Fewer/ no enzyme-substrate complexes form/ fewer successful collisions;

O Max. rate of reaction cannot be achieved/increasing the concentration of the substrate has no effect on inhibition; allow correctly labelled graph

6.

(a)

A – Biological catalysts that speed up the rate of reactions.

B – Lower the activation energy of a reaction.

C – (Very) specific due to tertiary / globular structure of protein.

D – Description of lock and key. Accept labelled diagram.

E – Formation of enzyme substrate complex. Accept labelled diagram.

F – Importance of active site.

G – Description of the induced fit theory.

H – pH alters structures / activity / work at optimum pH (not: denatures)

I – An increase in the concentration of substrate increases the rate of reaction, or converse.

J – An increase in the concentration of enzyme increases the rate of reaction, or converse.

K – Hydrogen bonds, disulphide bonds, van der Waals' forces Any 2 from 3

L – Enzymes can be used again as they are not affected by the reaction / are unchanged at the end of the reaction.

M – Enzymes can be denatured by high temperature as it changes the shape of the active site.

(A maximum of 10 marks can be awarded from the 14 available.) [10]

7.

(a) Enzyme essay mark scheme

a Biological catalysts/speed up a chemical reaction
(not: catalyst unqualified)

b By lowering activation energy/labelled diagram

c Globular protein/labelled diagram/tertiary structure or eq.

d With active site/specific shape

e Into which substrate(s) fits and is converted to product(s)

f Correct reference to lock and key/induced fit or suitably labelled diagrams

PH

g Enzymes have a narrow optimum pH range

h Large deviations from this optimum may result in denaturation/shape change

i Small pH changes cause reversible changes (which may inactivate the enzyme)

Temp

j Rate of reaction increases with increasing temperature

k Because of increased number of collisions of active site and substrate

l Enzymes are denatured by higher temperatures above 45°C
Optimum for most (mammalian) enzymes approx 37°C

m At low temperatures enzymes are inactive/eq

Inhibitors

n Description of the 2 types of inhibitors competitive

p Non-competitive/labelled diagrams

q Correct egs cyanide for non-comp, malonic acid for comp

Must mention pH, temp and inhibitors to get 10 marks. If one or more missing Max. 9 marks.

8.

(a)

A	Definition of inhibition – slowing or stopping of an enzyme's action by another substance.	1 mark
B	In competitive inhibition, molecule combines with active site.	1 mark
C	Because molecule with similar shape to the substrate.	1 mark
D	Level of inhibition depends on relative inhibitor to substrate concentrations	1 mark
E	End product inhibition	1 mark
F	Non competitive inhibitors combine with the enzyme at another point away from the active site.	1 mark
G	This distorts the shape of the enzyme protein causing alteration in the active site	1 mark
H	which prevents the formation of the enzyme-substrate complex	1 mark
I	The enzyme molecule may be permanently damaged.	1 mark
	(Five marks maximum)	
J	Axes correct plus 1 correct line	1 mark
K	Axe correct plus 3 lines correct	1 mark

L	Graph is horizontal when all active sites are occupied.	1 mark
M	Competitive plots shows slowing of rate but less effective as relative substrate concentration rises.	1 mark
N	Eventually relative inhibitor concentration is too low to affect many of the active sites (point x).	1 mark
O	Non competitive 'knocks out' fixed amount of enzyme.	1 mark
P	Horizontal level is lowered because there is effectively a lowered enzyme concentration.	1 mark
	(Five marks maximum for J-P; at least one of which must be for the graph i.e. J/K)	
		Total 10 marks

9.

A. Protein + reference primary structure ie sequence amino acids

B. Reference secondary structure- description and explanation;

C. 3D folding;

D. Tertiary / globular;

E. Reference hydrogen bonding / ionic bonding /

hydrophobic interaction;

F. Reference disulphide bridges;

G. Active site as (specific) binding site for substrate;

H. Binding (with active site) lowers activation energy;

I. Collision theory eg ref. temperature and more successful collisions

J. Heat denaturation explained ie change shape of active site

K. Effects pH ie change of active site

L. Consequences of changes to active site ie substrate no longer fits (J or K);

M. Reference to small change in pH reversible , high irreversible;

N. Reference competitive inhibitors binding to active site / co factors

O. Reference non competitive inhibition binding not at active site;

P. therefore changing shape of active site

Q. induced fit/ lock and key